International Journal of Drug Delivery 8 (2016) 125-133
http://www.arjournals.org/index.php/ijdd/index

Original Research Article

ISSN: 0975-0215

Formulation and *in vitro/in Vivo* evaluation of Olmesartan medoxomil solid dispersions incorporated E/R trilayer matrix tablets by geomatrix

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Abstract

An attempt has been made to develop and optimize an novel anti hypertensive trilayered controlled release matrix tablets incorporated with Olmesartan medoxomil solid dispersion prepared by direct compression and consisted of middle active layer with different grades of hydroxypropyl methylcellulose (HPMC), guar gum, ethyl cellulose. Upper and lower layers are prepared with Carnauba wax, guar gum and sodium CMC. The developed drug delivery system provided prolonged drug release rates over a period of 24 h. The release profile of the optimized formulation (HF14) was described by the Zero-order and Higuchi model. In-vivo bioavailability studies were carried out with the optimized formulation (HF14) and reference standard A fair correlation between the dissolution profile and bioavailability for the optimized formulation was observed. The results indicate that the approach used could lead to a successful development of a trilayer extended release formulation up to 24h. These results also demonstrated that the Olmesartan solid dispersion incorporated trilayer tablets shown more bioavailability because of its conversion from crystalline to amorphous form.

Keywords: Olmesartan medoxomil, Trilayer matrix tablet, Guar gum, Geomatrix, In-vivo bioavailability studies.

Introduction

Oral drug delivery has been known for decades as the most widely utilized route of administration among all the routes that have been explored for the systemic delivery of drugs via various pharmaceutical products of different dosage forms [1]. Controlled release pharmaceutical systems have been developed and studied to improve the performance of drugs and in particular to increase their pharmacological effect and reduce any side effects [2].

A number of design options are available to control or modulate drug release from a drug delivery system. Most oral controlled release dosage forms fall in the category of matrix, reservoir or multi-layer systems. Lately, multi-layer matrix systems are gaining importance in the design of oral sustained drug delivery systems. A multi-layer system consists, usually, of a hydrophilic matrix core containing the active ingredient and one or two impermeable or semi-permeable polymeric coatings (barrier-layer) applied on one or both faces of the core during tabletting [3-5]

The barrier layers delay the interaction of active solute with dissolution medium, by limiting the surface available for the solute release and at the same time controlling solvent penetration rate [6,7]. In the device, the coat layers prevent the water penetration through the protected core for some duration. After this phase during the subsequent dissolution process, the swollen barriers erode and the surface available for drug release slowly increases. In this way the decrease of delivery rate due to the increase in diffusion path length is counter balanced by the simultaneous increase of the area available for drug release [8,9] The use of naturally occurring biocompatible gums has been the focus of recent research activity in the design of dosage forms for oral controlled release administration, and hydrophilic polymers matrix systems are widely used because of their flexibility to provide a desirable drug release profile, cost effectiveness, and broad regulatory acceptance [10].

There have been different approaches to achieve zero-order drug release from dosage forms for sustained plasma concentration. Among different approaches to achieve zero-order release from hydrophilic matrix technologies, multilayer matrices have been widely evaluated and developed for commercial products under the trade name of Geomatrix. The technology makes use of bilayer or trilayer tablets to modulate the release and to achieve constant release [11].

Olmesartan Medoxomil is a non peptide angiotensin II receptor antagonist use in the treatment of mild to moderate hypertension. This drug is weakly basic, lipophilic and having very less oral bioavailability of about 26%. Olmesartan Medoxomil inhibits type I angiotensin II receptor in the rennin angiotensin system, there by producing best antihypertensive action [12].

The main aim of the present study was to prepare Olmesartan solid dispersions to enhance its water solubility as it belongs to BCS class II and bioavailability (low bioavailability of 26%) and it was incorporated in trilayered controlled release matrix tablets with

DOI:10.5138/09750215.1948

different hydrophobic and hydrophilic polymers to achieve zero-order drug release for sustained plasma concentration.

Materials and Methods

Materials

Olmesartan pure drug was generous gift from Aurobindo Pharma Ltd., Hyderabad, India. Sodium carboxyl methyl cellulose, Ethyl cellulose, HPMC K 4 M, HPMC K & HPMC K 100 M was obtained from Rubicon labs, Mumbai. Guar gum was gifted from MSN Labs Ltd. Hyderabad. All other chemicals used were of analytical grade.

Methods

Formulation of controlled release Olmesartan trilayer matrix tablets

The trilayered matrix tablets of Olmesartan were prepared by direct compression method. The first step in the formulation was to develop the middle active layer so as to give at least 90%drug release during 12hours. This layer would then be sandwiched between barrier layers (Upper & Lower layers) so as to continue the drug release for 24hours.

Preparation of middle active layer

Fourteen formulations (F1-F14) for active layer were formulated using polymers like different grades of HPMC (HPMC K14M & HPMC K100M) and Guar gum. All the formulations were varied in concentration of release retardant polymers and Olmesartan solid dispersions [13], talc (1.5mg) & magnesium stearate (1.5mg) constituted in all the formulations. These materials were screened through 60 and mixed together in motor by using pestle. Final mixtures were compressed by using 12mm diameter flat punches on a sixteen station rotary tablet press where only one station was operative and other station were nullified Formulation F1-F14 containing drug and other polymers prepared under condition as showed in table.

Table 1: Formulation trails for active layer (F1-F7)

INGREDIENTS (mg)	F1	F2	F3	F4	F5	F6	F7
Olmesartan Medoxomil (Solid dispersion)[13]	160	160	160	160	160	160	160
HPMC K 4 M	20	25	30	32	35	40	45
HPMC K 100M	---	---	---	----	---	----	----
Ethyl cellulose	25	23	22	21	21	20	18
Guar gum	15	15	10	12	12	09	09
Sodium carboxy methyl cellulose	12	11	11	9	10	09	08
Dibasic calcium phosphate	15	13	14	13	09	09	07
Magnesium stearate	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Talc	1.5	1.5	1.5	1.5	1.5	1.5	1.5

Table 2: Formulation trails for active layer (F8-F14)

INGREDENTS (mg)	F8	F9	F10	F11	F12	F13	F14
Olmesartan medoxomil (SD)	160	160	160	160	160	160	160
HPMC K 4M	---	----	----	----	----	----	----
HPMC K 100M	25	30	35	37	40	45	49
Ethyl cellulose	12	20	15	15	17	12	09
Guar gum	09	12	16	11	9	15	10
Sodium carboxy methyl cellulose	06	15	13	12	11	11	10
Dibasic calcium phosphate	06	15	13	14	13	09	09
Magnesium stearate	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Talc	1.5	1.5	1.5	1.5	1.5	1.5	1.5

Preparation of barrier layers

The barrier layers (Upper & Lower layers) was formulated employing hydrophobic polymers Carnauba wax and Guar gum, which include water soluble DCP & EC. Composition of barrier layers was depicted in Table 3.

The procedure tried to make the compacts was via direct compressions. For the first procedure the carnauba wax, xanthan gum and the filler was mixed in mortar and lubricated with magnesium stearate. The mix is then compressed using rotary press having 12mm flat tooling.

Table 3: Formulation trails for barrier layer

Ingredients (mg)	A	B	C	D	E	F	G	H
Carnauba wax	5	10	15	20	25	30	35	40
Guar gum	22	24	22	18	20	22	20	20
Ethyl cellulose	20	16	20	17	18	10	12	12
Dibasic calcium Phosphate	50	47	40	42	34	35	30	25
Magnesium stearate	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Talc	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5

Formulation of Olmesartan tryilayer tablets

The powder mixtures required for active and barrier layers were weighed accurately and thoroughly mixed using mortar and pestle for about 20 minutes. Initially, the volume of die cavity (12mm, round) was adjusted equivalence to the weight of trilayered matrix tablets (450mg). Then the pre weighed amount of powder equivalent to bottom layer (100mg) was taken and placed in the die cavity and slightly compressed for uniform spreading. The upper punch was lifted up and 250mg of the drug containing middle active layer optimized formulation (F14) was placed over the bottom layer in the die cavity and again slightly compressed. The remaining volume of the die cavity was filled with pre weighed (100mg) amount of powder equivalent to top layer and compressed with the full force of compression on rotary tablets press to obtain tri-layered tablets. Tri-layered matrix tablets of each composition were compressed and tested for their friability, Hardness, drug content and drug release characteristics with a suitable number of tablets for each test.

Table 4: Composition of Olmesartan trilayer matrix tablets

INGREDIENTS (mg)	AF14	BF14	CF14	DF14	EF14	FF14	GF14	HF14
MIDDLE LAYER (F14)								
Olmesartan	160	160	160	160	160	160	160	160
HPMC K 100M	49	49	49	49	49	49	49	49
Ethyl cellulose	09	09	09	09	09	09	09	09
Guar gum	10	10	10	10	10	10	10	10
Sodium CMC	10	10	10	10	10	10	10	10
Dibasic calcium phosphate	09	09	09	09	09	09	09	09
Magnesium stearate	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Talc	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
BARRIER LAYER (on each side)								
Carnauba wax	5	10	15	20	25	30	35	40
Guar gum	20	16	20	17	18	10	12	12
Ethyl cellulose	22	24	22	18	20	22	20	20
Dibasic calcium Phosphate	50	47	40	42	34	35	30	25
Magnesium stearate	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Talc	1.5	1.5	1.5	1.5	1.5	1.5	1.5	1.5

Evaluation of Olmesartan trilayer matrix tablets

Weight variation, hhardness, thickness, ffriability were evaluated.

Drug content / Assay

20 tablets were accurately weighed and powdered.10mg equivalent powder was dissolved in 50ml distilled water and sonicated for 15 minutes. It was filtered and washed with distilled water. Filtrate and washings were combined. Final volume was made up to 100ml with distilled water. Absorbance of this solution was determined in a UV spectrophotometer at 248nm. Amount of Olmesartan in tablets was calculated by using regression equation.

In-vitro drug release profile

In vitro drug release studies for developed Olmesartan trilayer matrix tablets were carried out by using dissolution apparatus II paddle type (Electrolab TDL-08L). The drug release profile was studied in 900ml Phosphate buffer pH 6.8 at 37± 0.5^0C temperature. The amount of drug release was determined by UV visible spectrophotometer (Shimadzu UV 1800) at 248nm.

Drug release kinetics

To describe the kinetics of the drug release from matrix tablet, mathematical models such as Zero-order, First order and Higuchi, models were used. The criterion for selecting the most appropriate model was chosen on the basis of the goodness-or fit test.

Drug-excipient compatibility studies

Fourier transform infrared spectroscopy (FTIR)

FTIR spectra for pure drug, physical mixture and optimized formulations were recorded using a Fourier transform Infrared spectrophotometer. The analysis was carried out in Shimadzu-IR Affinity 1 Spectrophotometer. The samples were dispersed in KBr and compressed into disc/pellet by application of pressure. The pellets were placed in the light path for recording the IR spectra. The scanning range was 400-4000 cm^{-1} and the resolution was 1 cm^{-1}.

Pharmacokinetic study of Olmesartan medoxomil

Animal Preparation

Male rabbits were (weighing 2-3kg) selected for this study, all the animals were healthy during the period of the experiment. Animals were maintained at room temperature 25^0C, Relative Humidity 45% and 12h alternate light and dark cycle with 100 % fresh air exchange in animal rooms, uninterrupted power and water supply and rabbits were fed with standard diet and water ad libitum. The protocol of animal study was approved by the institutional animal ethics committee Vijaya College of Pharmacy, Munganoor, Hyathnagar, Rangareddy dist.(IAEC NO: P40/VCP/IAEC/2015/9/DBP/AE15/Rabbits).

In vivo study design

The rabbits were randomly divided into two groups each group contains six animals. The group A was received prepared Olmesartan matrix tablets (40mg), standard conventional tablets (40mg) was administered group B with equivalent dose of animal body weight. Blood samples (approximately 0.5ml) were obtained with syringes by marginal ear vein at 0, 0.5, 1, 1.5, 2, 4, 6, 8, 12, 16, 20 and 24h post dose. During collection, blood sample has been mixed thoroughly with heparin in order to prevent blood clotting. Plasma was separated by centrifugation of the blood at 5000rpm in cooling centrifuge for 5min to 10min and stored frozen at 20 C until analysis.

Preparation of Plasma Samples for HPLC Analysis

Rabbit plasma (0.5 ml) samples were prepared for chromatography by precipitating proteins with 2.5 ml of ice-cold absolute ethanol for each 0.5ml of plasma. After centrifugation the ethanol was transferred into a clean tube. The precipitate was re suspended with 1 ml of Acetonitrile by vortexing for 1 min. After centrifugation (5000 – 6000rpm for 10min), the Acetonitrile was added to the ethanol and the organic mixture was taken to near dryness by a steam of nitrogen at room temperature.

HPLC Method

C18 column with 5µm particle size and the Mobile Phase consisting of binary mixture of 10mM mixed phosphate buffer (pH 3.0 ± 0.05) and methanol in a ratio of 40:60v/v. The flow rate was 1.0ml/min and the effluents were monitored at 255nm. Internal standard Eprosartan was used. The retention times was 5.15min and 2.87min respectively Olmesartan and Eprosartan [14].

Pharmacokinetic analysis

The pharmacokinetic parameters employed to evaluate were maximum plasma concentration (C_{max}), time to attain C_{max} i.e., T_{max} and $t_{½}$ values, area under plasma concentration–time curve from zero to the last sampling time (AUC_{0-t}), area under plasma concentration–time curve from zero to infinity (AUC_{0-}). AUC_{0-t} was calculated by the linear trapezoidal rule and AUC_{0-} from the following formula.

$$AUC_{0-} = AUC_{0-t} + C_t / K_E$$

Results and Discussion

Preparation of Olmesartan active layer

The matrix tablets of Olmesartan were prepared without the barrier layers by direct compression method. All the formulation trails were subjected to *in vitro* dissolution to determine the drug release profile.

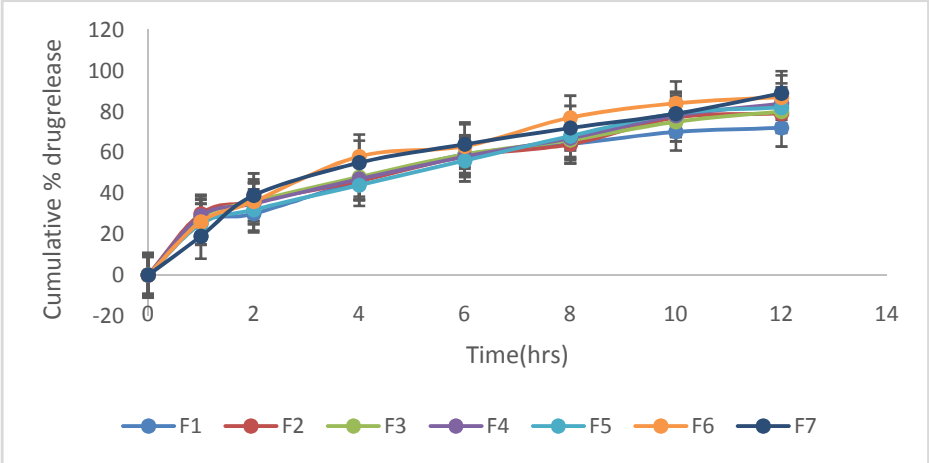

Figure 1: In vitro Dissolution profile of F1-F7 Olmesartan active layer formulations

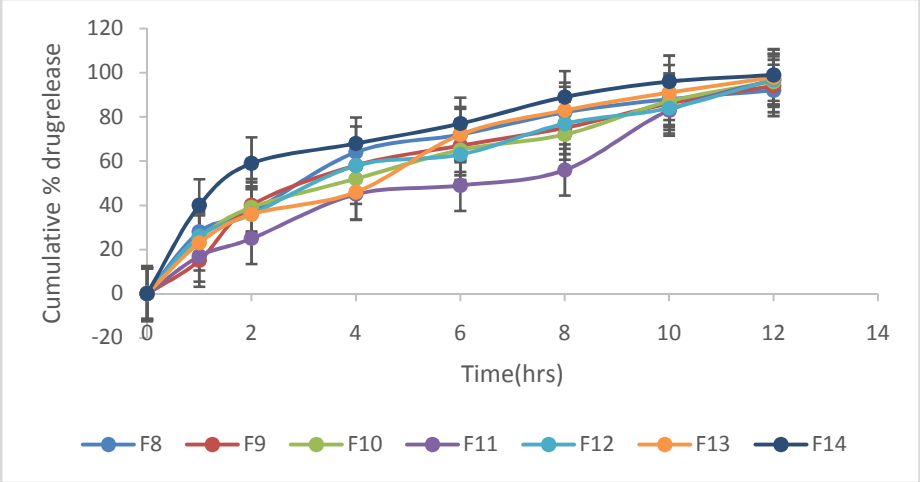

Figure 2: In vitro Dissolution profile of F8-F14 Olmesartan active layer formulations

From the above results, the formulation F14 was decided as optimized formulation based on the highest drug release i.e. 99.2±0.10 upto 12 h when compare with other formulations as active layer of the trilayer tablets.

Table 5: Evaluation parameters of Olmesartan trilayer matrix tablets

Formulation code	Thickness (mm)	Hardness (kg/cm^2)	Friability (%)	Weight variation (mg)	%Drug content
AF14	4.64	6.6±0.23	0.27	448±12	98.4
BF14	4.71	7.1±0.42	0.23	447±11	96.9
CF14	4.54	7.4±0.41	0.25	448±16	97.2
DF14	4.73	7.3±0.32	0.25	447±14	96.7
EF14	471	7.4±0.59	0.28	448±11	98.8
FF14	4.68	6.7±0.22	0.26	446±16	96.7
GF14	4.55	7.6±0.55	0.24	448±13	98.4
HF14	4.72	7.1±0.15	0.21	449±16	99.2

The evaluation parameters of all the tablets are within the limits and the hardness ranges in between 6-7kg/cm^2. The percentage drug content was between 96-99. The Friability, weight variation and thickness was found to be within the limits. Hence all the tablets were subjected to in vitro dissolution test to determine the release profiles.

Table 6: *In-vitro* dissolution studies of Olmesartan trilayer tablets

Time (h)	AF14	BF14	CF14	DF14	EF14	FF14	GF14	HF14	Reference standard
1	12.34±0.01	14.16±0.01	16.21±0.04	18.22±0.04	19.22±0.04	20.85±0.04	20.47±0.05	21.02±0.04	96.01±0.06
2	22.11±0.02	24.28±0.02	22.23±0.05	23.21±0.05	25.32±0.02	21.25±0.05	24.54±0.05	26.99±0.09	
4	32.15±0.13	33.38±0.34	34.28±0.05	36.32±0.04	36.35±0.03	37.23±0.45	38.43±0.05	39.23±0.03	
6	41.25±0.05	42.45±0.03	43.79±0.05	44.15±0.14	45.62±0.04	46.54±0.05	48.25±0.04	49.99±0.05	
8	51.16±0.19	52.98±0.094	53.23±0.05	55.42±0.08	56.32±0.05	57.15±0.04	58.45±0.02	59.99±0.07	
12	60.34±0.05	62.99±0.05	53.75±0.06	64.12±0.05	66.46±0.05	66.99±0.05	68.74±0.03	69.97±0.05	
16	69.75±0.43	72.55±0.11	73.68±0.26	75.24±0.24	76.53±0.14	78.32±0.01	78.65±0.03	79.99±0.05	
20	80.24±0.05	81.20±0.05	82.95±0.07	84.21±0.05	85.15±0.06	88.22±0.05	86.85±0.02	89.55±0.06	
24	86.12±0.04	90.24±0.03	93.45±0.01	91.22±0.02	93.55±0.04	94.24±0.04	95.25±0.02	98.56±0.19	

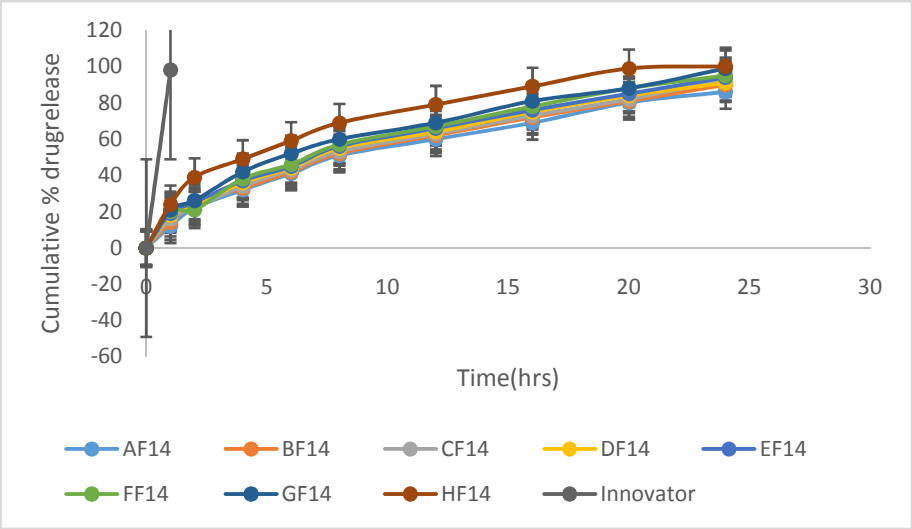

Figure 3: Comparison of Cumulative percentage drug release of Olmesartan trilayered matrix tablets and reference standard.

The *In vitro* drug profile of Olmesartan from different formulations was carried and the results are depicted in Table 6 & Figure 3. The trilayer tablets extended the drug release up to 24hrs. The highest drug release was found in the formulation HF14 i.e 98.56±0.19% within 24hrs. HF14 was found to be optimized formulation based on the dissolution and other evaluation parameters. The in vitro drug release profile from reference standard conventional tablet was found to be 96.01±0.06% within 60min.

Release order kinetics of Olmesartan trilayer matrix tablets with reference standard

In the present study drug release mechanism of Olmesartan trilayer matrix tablets are best fitting to zero order and Higuchi model because regression coefficient was seen closest to 1 in these models which conforms diffusion assisted mechanism of release. The reference standard release was explained by first order kinetics as the plot showed highest linearity as the drug release was best fitted in first order kinetics. The results are summarized in Table.

Table 7: Release order kinetics of Olmesartan trilayer matrix tablets with reference standard.

Formulation code	Zero order R^2	First order R^2	Higuchi R^2	Korsmeyer Peppas R^2	Korsmeyer Peppas n value
AF14	0.979	0.862	0.946	0.975	0.558
BF14	0.980	0.863	0.947	0.976	0.604
CF14	0.984	0.867	0.951	0.981	0.573
DF14	0.982	0.865	0.949	0.979	0.634
EF14	0.985	0.868	0.952	0.982	0.598
FF14	0.987	0.869	0.954	0.983	0.721
GF14	0.989	0.870	0.955	0.985	0.719
HF14	0.991	0.871	0.957	0.987	0.695
Reference standard	0.916	0.977	0.928	0.905	0.665

Characterization FT-IR studies

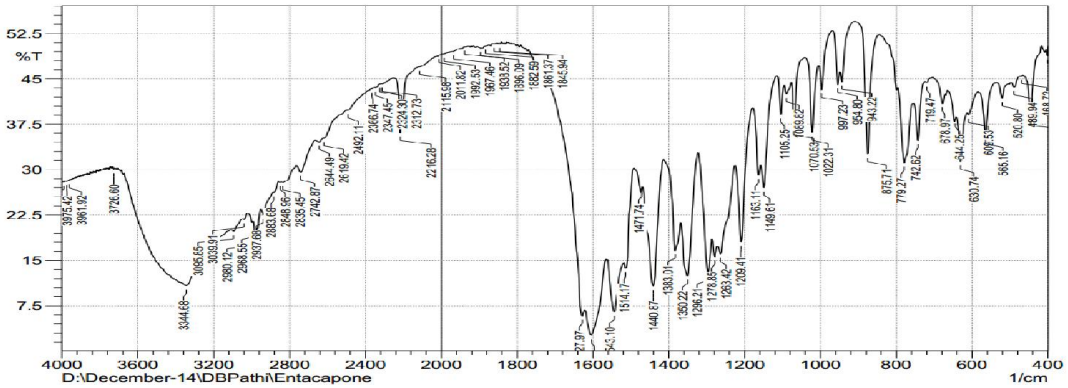

Figure 4: FT-IR spectrum of pure drug Olmesartan medoxomil

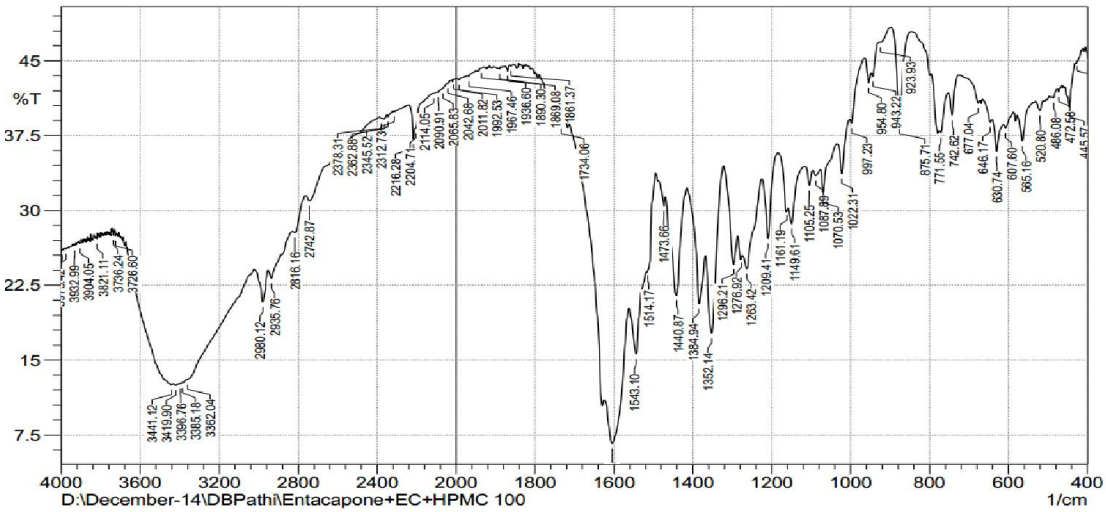

Figure 5: FT-IR spectrum of physical mixture

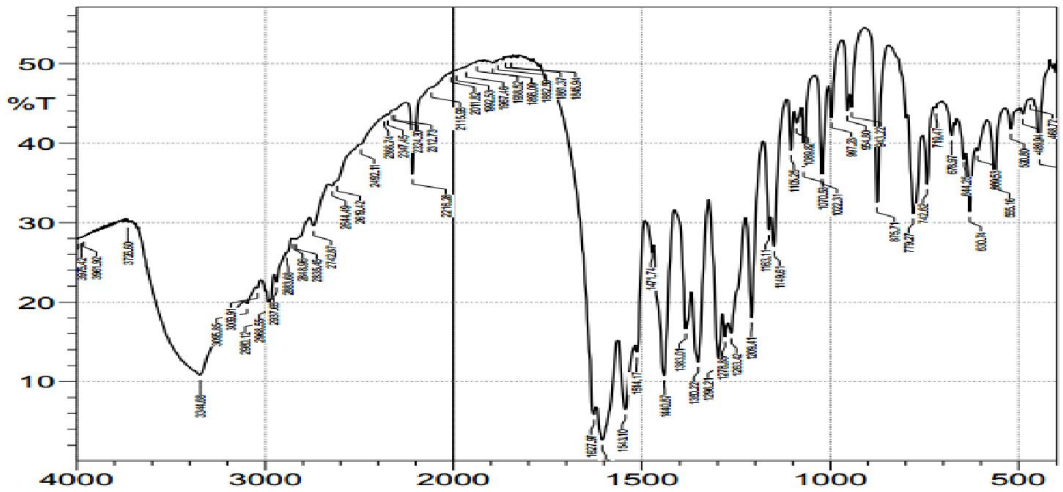

Figure 6: FT-IR spectrum of optimized formulation HF14

From FTIR spectrum there was no alteration in peaks of Olmesartan pure drug and optimized formulation, suggesting that there was no interaction takes place between drug & excipients.

The FTIR spectrum of Olmesartan pure drug, with physical mixture and optimized are shown in Figure 4, 5 & 6 respectively.

Pharmacokinetic studies

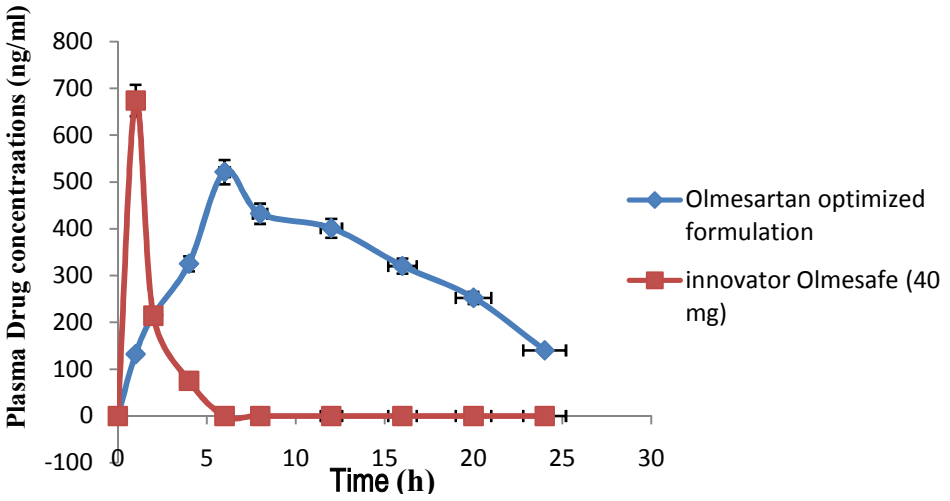

Figure 7: Plasma Concentrations of Olmesartan Optimized formulation and Reference standard at different time intervals

Table 8: Comparison of pharmacokinetic parameters of Olmesartan Optimized formulation and Reference standard

Parameters	Olmesartan Optimized formulation	Reference standard
C_{max} (ng/ml)	521.11±0.05	674.11±0.01
AUC $_{0-t}$ (ng h/ml)	7135.65±0.12	3523.25±0.02
AUC_{0-} (ng h/ml)	8167.75±0.14	3945.14±0.02
T_{max} (h)	6.02±0.14	1.15±0.12
$t_{1/2}$ (h)	7.5±0.014	2.50±0.05

Bioavailability Parameters

Mean plasma concentration profiles of prepared Olmesartan optimized formulation and marketed product are presented in Figure 7. Olmesartan optimized formulation exhibited as sustained release in vivo when compared with marketed tablet. All the pharmacokinetics parameters displayed in Table 8. In this study the prolonged drug absorption was achieved with the test formulation. The average peak concentration of the reference formulation was higher than that of the test (674.11±0.01ng/ml for the test formulation versus 521.11±0.05ng/ml for the reference). AUC is an important parameter in evaluating bioavailability of drug from dosage form, as it represents the total integrated area under the blood concentration time profile and represents the total amount of drug reaching the systemic circulation after oral administration. $AUC_{0\text{-}inf}$ for optimized formulation was higher (8167.75±0.14 ng h/ml) than the reference standard 3945.14±0.02ng h/ml. Statistically, $AUC_{0\text{-}t}$ of the optimized preparation was significantly higher ($p<0.05$) as compared to reference standard. Higher amount of drug concentration in blood indicated better systemic absorption of Olmesartan from optimized formulation as compared to the reference standard product.

Summary and Conclusion

All carnauba wax and guar gum formulations displayed sustained release with the Olmesartan incorporated solid dispersion trilayer matrix tablets, however three-layer tablet formulations demonstrated lower drug release compared to matrix tablets. Based on the evaluation parameters, drug dissolution profile and release drug kinetics HF14 was found to be optimized formulation. The drug release from HF14 was found to fit Zero order and best fitted to Higuchi's model confirming to be diffusion assisted mechanism. FTIR studies revealed that, there was no interaction between the drug and polymers used in the formulations. In vivo bioavailability studies were conducted for optimized formulation HF14 and reference standard. The optimized formulation of Olmesartan trilayer matrix tablet was shown significant plasma concentration with controlled release and maintained for 24 hrs.

Conflict of Interest

The authors declare no conflict of interest.

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